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<b>(54) Title:</b> MODIFIED FOOD PRODUCTS AND BEVERAGES, AND ADDITIVES FOR FOOD AND BEVERAGES  <b>(57) Abstract</b>  A method for making a composition suitable for inclusion in a food product or beverage the method comprising the step of combining a hydrophobic compound which can be shown to be beneficial for human health with a component which is acceptable as a food additive, wherein the component which is acceptable as a food additive interacts with the surface of the hydrophobic compound. Preferably, the hydrophobic compound is a plant sterol or lycopene or a combination thereof. Food products and beverages supplemented with plant sterol and other hydrophobic compounds are provided. In particular, the food products or beverages are an emulsifiable spread or ones which are fermented with lactic acid bacteria.		

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## **MODIFIED FOOD PRODUCTS AND BEVERAGES, AND ADDITIVES FOR FOOD AND BEVERAGES**

### **FIELD OF THE INVENTION**

5           The present invention relates to modified food products and beverages, particularly those containing plant sterols, and it relates to additives for food and beverages, particularly plant sterol additives.

          The application describes novel plant sterol containing additives, and a method for their preparation and their general use in food products  
10   and beverages; food products which are produced by the fermentation of a mixture of raw material and plant sterols with lactic acid bacteria and, more specifically to fermented milk products such as yoghurt; emulsified fat products, and more specifically, to low fat spreads, which contain the novel plant sterols additives of this invention; and processes for the  
15   preparation of the above said food products and beverages, which are supplemented with plant sterols. In addition, the application describes novel additives which contain other hydrophobic compounds which are known to benefit human health, such as lycopenes, and their use in food products and beverages.

20

### **BACKGROUND OF THE INVENTION**

          The medicinally useful properties of plant sterols have been known for a considerable time. In 1953 Pollak reported the preventive effect of

plant sterols on experimental atherosclerosis in rabbits (*Circulation*, (1953) 7, 696-701) and plant sterols-induced reduction of the level of blood serum cholesterol in man (*Circulation* (1953) 7, 702-706). Three years later Farquhar *et al* (*Circulation*, (1956) 14, 77-82) demonstrated a significant  
5 reduction of serum cholesterol in young men with atherosclerotic heart disease. Other therapeutic effects e.g. in the treatment and prevention of diverse geriatric diseases, hypertrophy of the prostate, rheumatic and hyperlipidaemic manifestations, hyperglycaemia as well as anti-inflammatory and haemostatic actions, were also described for various  
10 modified sterols such as saponins (US Patents 4 602 003 & 4 242 502), dihydrocholanes (US Patent 4 117 121) and sterol glycosides and their derivatives (British Patents 1 298 047 & 1 491 532, US Patents 3 991 186, 4 188 379 & 4 254 111).

The mechanism of action of plant sterols is not completely  
15 understood at present. Also, certain controversy exists with regard to the relative potency of individual sterols and their derivatives and the most efficient way of administering them. Thus, Ikeda and Sugano (*Atherosclerosis*, (1977) 30, 227-237) basing their conclusion on animal trials suggested that stanol, which is the 5 $\alpha$ -saturated derivative of  
20 sitosterol, has somewhat higher hypocholesterolemic activity than sitosterol. Miettinen *et al* (US Patent 5 502 045) reported that stanol esters are more potent in lowering cholesterol than unesterified stanol and suggested that the difference was due to better solubility of the former

compounds in the oil matrix which was used to administer them. On the other hand, Mattson *et al* (*Am. J. Clin. Nutr.*, (1982) **35**, 697-700) found the opposite effect when comparing the efficacy of sitosterol suspension and oil soluble sitosterol oleate, whilst in another study (*J. Nutr.* (1977) **107**, 1139-1146) no appreciable difference between free sterols and their esters was found. The recent extensive study by Weststrate and Meijer (*Eur. J. Clin. Nutr.*, (1998) **52**, 334-343) also suggests that the esters of sitosterol and stanol esters, when tested in parallel trials, are equally effective in lowering the level of serum cholesterol in humans. However, irrespective of the relative potency of various plant sterols and their derivatives, it has been well established in the prior art that these compounds lower the level of serum cholesterol in humans. It is also clear that laborious and expensive modifications of plant sterols by, for example, hydrogenation (WO 98/38206) or esterification (WO 98/01126) are unnecessary and are not necessarily in the interest of the consumer who will have to, at the end of the day, pay the costs associated with these modifications.

Due to their beneficial effects, and the fact that plant sterols are naturally occurring components of vegetable fats and oils (and hence their consumption is considered to be safe), plant sterols were also suggested for use in food formulations by, for example, dissolving sterols in an effective food-acceptable solubilising agent such as triglycerides, vegetable oils, tocopherols, polyols and alcohols (US Patent 5 244 887;

EP 0 839 458; EP 0 289 636; WO97/42830; WO 98/31372; ). Other vegetable oil-based products, notably margarine, mayonnaise and fat spreads which contained plant sterols and their esters, were disclosed in several patent applications (US Patent 5 502 045; WO 97/42830; WO 5 98/06405; WO 98/01126; WO 98/19556). At least one of these products (US Patent 5 502 045) has been approved for human consumption and margarine supplemented with stanol esters is now available on the market under the tradename Benecol. Thus, it is generally accepted that plant sterols and their esters in dosages of about 1 gram per day or more can 10 reduce the level of blood cholesterol and that they are generally well tolerated in long term use.

It is well known that plant sterols are very poorly soluble in water (The Merck index, 11th Edition, 1989, Ed. S. Budavari). Consequently vegetable oil-based formulations have been often used in the prior art for 15 the incorporation of plant sterols in food products. However, these foods are not suitable for those consumers who try to limit their fat intake due to various health reasons and/or specific dietary requirements. There is therefore a need in the trade to develop and introduce new products supplemented with plant sterols, including low fat and non-fat or virtually 20 fat free products, to satisfy consumer demand. It is also clear from the prior art that in order to draw the maximum benefit from such products, they should be consumed on a regular basis and in sufficient quantities. Emulsified low fat spreads, fermented dairy products and some other

common foods and beverages described herein satisfy these criteria.

Fat spreads and margarine can be prepared with different fat content as legally specified and typically between 10% and 80% fat by weight and the products can be labelled accordingly as, for example, low fat or very low fat spreads. The latter are especially appealing to many health conscious consumers. Generally, depending on the fat content and other ingredients used, a fat spread is a water-in-oil or oil-in-water emulsion (or a combination of the two) which has a butter like consistency and taste and which is spreadable. A number of such spreads which are supplemented with plant sterols, and methods for their manufacture are known in the prior art. For example, a pharmaceutically stable emulsion of finely dispersed plant sterols is disclosed in US 3 085 939; US 4,195,084 and EP 0 289 636. According to these inventions chemically unmodified plant sterols are used and they are contained predominantly in the aqueous phase of the product. However, the dispersion according to these invention is fluid and therefore it does not have a taste and consistency like butter and is not spreadable. On the other hand, the products produced according to US 5,502,045, WO 98/06405, WO 98/19556 and WO 98/01126, satisfy the requirements i.e. have a taste and consistency like butter and are spreadable but they contain plant sterols which were subjected to chemical modifications and the sterols are incorporated into the oil phase of the product in the form of esters. Thus, according to these inventions an expensive and unnecessary chemical

modification of plant sterols is required prior to their incorporation into a fat spread.

Some processes of incorporating unmodified plant sterols into fat spreads are also known in the prior art. For example, Tianen *et al* (WO 5 98/13023) disclosed the process of adding plant sterols to a pre-prepared (i.e. already emulsified) fat spread by admixing finely ground sterols with the spread. However, according to this invention an expensive grinding treatment of plant sterols to reduce the particle size to about 30 micron is necessary to improve the organoleptic properties of this product.

10 Generally the poor organoleptic properties of this and similar products result from a different organoleptic perception of the particulate sterols in the oil and water phases between which the sterol powder is evenly distributed by admixing. Also the process of this invention is incompatible with most modern methods of manufacturing fat spreads which are often

15 packed into tubs straight after emulsification. The process of this invention cannot be readily used for the production of low and very low fat spreads as the texture of these spreads and their appearance will be adversely affected by blending with particulate material. Therefore there is a need in the trade to produce a fat spread, and especially a low and a very low fat

20 spread, which contains plant sterols and have acceptable organoleptic properties. There is also a need in the trade to develop a suitable sterol-containing additive which use would be compatible with the current manufacturing methods for the production of such spreads. These needs



are addressed by the present invention.

Yet another group of foods and beverages which since ancient times have formed an important part of the human diet in many regions of the world are fermented food products and beverages. Although such foods vary widely in their appearance, texture and organoleptic properties in accordance with the raw materials used for their production, they typically share several common features. Firstly, their production involves at least three common steps: (1) pasteurisation of the raw material; (2) contacting the pasteurised material with a specific micro-organism or a mixture of micro-organisms and (3) incubating the mixture of raw material and micro-organisms for sufficient time to enable the fermentation to occur. Secondly, these fermented foods typically contain micro-organisms, dead or alive, as well as metabolites of their fermentation which are believed to be beneficial to human health. Thirdly, many of these foods constitute a substantial part of the diet and are consumed on a regular basis.

Rice and soybeans are the most common raw materials for the production of fermented foods in many Eastern countries, including Japan. Soybeans, for example, are fermented to produce Japanese "miso" (soybean paste), "tofu" (soybean protein curd) and many others. Soybean milk is also fermented to produce nutritionally valuable products.

In Western countries fermented milk products are most widely used. Examples of these products include yoghurt, sour cream, kefir, ymer as

well as fresh cheeses such as quark and fromage frais. Buttermilk, which is obtained as a by-product in the production of butter, can also be fermented using specific micro-organisms. The micro-organisms which are used in the production of fermented milk products, are known as  
5 starter cultures or starters. These micro-organisms are often mixtures containing lactic acid bacteria.

The nature of the product is determined to a considerable extent by the micro-organisms used which, depending on the species or strain, contribute to various degree to the formation of acid such as lactic acid,  
10 flavouring substances such as diacetyl or acetaldehyde and structure forming constituents such as polysaccharides. In some cases, one particular species of bacteria is used (such as *Lactobacillus acidophilus* in the production of acidophilus milk) but usually the product acquires characteristic properties as a consequence of the action of a mixture of  
15 micro-organisms on the raw material. For example, in kefir various other bacteria and yeast were detected in addition to *Lactobacilli* and *Streptococci*.

In many countries, notably Europe and USA, yoghurts are the most popular among fermented milk products. Yoghurt (as well as some other  
20 fermented milk products) contains an active culture of micro-organisms which can pass unharmed through the stomach. When the yoghurt micro-organisms enter into the intestinal area, they are believed to cause a favourable floral implantation and suppress undesirable bacteria in the

human digestive system. Yoghurt has also been found to be acceptable in the diet of those people who have malabsorption of lactose or an intolerance to lactose. Yoghurt is also believed to be beneficial to health due to its relatively high content of vitamins, minerals and other substances of considerable nutritional value. Consequently, yoghurt is consumed daily by a large proportion of the population in many countries as it is considered to be a healthy form of dairy product.

Yoghurt is generally made by a process that includes the following steps:

- (1) Yoghurt base mix which contains milk and added milk ingredients is pasteurised using a high temperature;
- (2) The pasteurised mix is then cooled down to a predetermined temperature;
- (3) A starter culture consisting of one or more yoghurt-producing micro-organisms is added (inoculation) and blended in while maintaining the lower temperature;
- (4) The fermentation is carried out until sufficient acid is produced and a thickened yoghurt product of the desired consistency (coagulum) is obtained;
- (5) The bacterial growth and metabolism are essentially arrested by cooling the yoghurt composition, usually to about 0°C-5°C.

Yoghurt can be prepared with a range of fat contents. For example, according to FDA, a yoghurt can be labelled as "non-fat", if it contains less

than 0.5% milkfat and as "low fat", if it contains from not less than 0.5% to no more than 2.0% milkfat. The standards and labelling practices vary from country to country. For example, non-fat or very low fat yoghurt in the UK should also have fat content below 0.5%, whilst in Germany and  
5 Australia the corresponding values are below 0.3% and below 0.2% respectively.

Depending upon the dairy ingredients employed, other ingredients and the processing treatment and methods, yoghurt may be prepared in various forms. Frozen yoghurt, at one extreme, is generally consumed in  
10 hard frozen or soft serve form. Liquid yoghurt, at the other extreme, is consumed by drinking, either directly or through a straw, rather than using a spoon. Usually yoghurts are classified according to their physical state. One of the most common form of yoghurt which is known as "firm" or "set type" is firm bodied, smooth and viscous, generally having the consistency  
15 of a light gel. Set yoghurt is filled into serving size cups, also known as retail containers, after the inoculation step and the containers are incubated at a suitable temperature regime. "Stirred" yoghurt is inoculated and incubated in a fermentation vessel and, when the fermentation is completed, the coagulum is broken during the cooling and packaging  
20 steps.

Fruits and flavouring agents are often added to yoghurt to suit particular tastes. Fruit-flavoured yoghurts are made Sundae style with fruit product at the bottom, Swiss style with fruit product pre-mixed, or Western

style with the fruit product at the bottom and the yoghurt is often additionally coloured and/or flavoured with natural and/or artificial colouring and flavouring agents. Fruited yoghurts are conventionally made with standard fruit products, also referred to as yoghurt fruit. Fruit  
5 products are generally made by cooking a fruit and sugar mixture with the addition of other agents such as colouring agents and thickeners. The cooked fruit product may contain discernible pieces of fruit in it or may be finely divided (puree form).

Sundae style fruited yoghurt is generally made in retail containers,  
10 by filling inoculated yoghurt mix over or under a suitable amount of fruit product and incubating the filled container for a suitable time. Swiss style fruited yoghurt is generally made by mixing a suitable amount of fruit product, after completion of the fermentation step. Fruited yoghurts typically contain from about 10% to about 30% fruit product by weight of  
15 the fruited yoghurt.

Drinking yoghurt is based on the stirring manufacturing process, except that a milk with lower solids content is generally used. The coagulum is broken down before filling into retail containers and fruit juice may be used instead of fruit product.

20 The shelf life of yoghurt can be increased by the so-called thermisation process which requires that the yoghurt is pasteurised and aseptically filled and the fruit product either added before the yoghurt or separately in its own container within the overall package. This process is

designed to increase the shelf life of the product but it also results in the destruction of beneficial micro-organisms in the yoghurt.

In the present specification all the described types of yoghurt will collectively be referred to as "yoghurt".

5

## SUMMARY OF THE INVENTION

This invention relates to the provision of novel food additives containing hydrophobic compounds which can be shown to be beneficial to human health, such as plant sterols and lycopenes (such as tomato lycopenes), and a method for their preparation and their use in various food products, beverages and processes involved in making food products and beverages. The method comprises coating the hydrophobic compound with food additives which are compatible with the above said products and processes, and in a preferred embodiment with milk-derived ingredients and proteins. A number of suitable coating agents are disclosed. Hydrophobic compounds which can be shown to be beneficial to human health also include carotenoids other than lycopene and poorly water soluble anti-oxidants.

This invention also relates, in one embodiment, to the discovery that by combining the hydrophobic compound (such as plant sterol or lycopene) with milk, milk-derived solids or other proteins in an aqueous phase of an emulsified fat spread prior to emulsification, the undesirable sensation in the mouth is substantially reduced or eliminated altogether

and a fat spread with good organoleptic properties is produced. According to this embodiment of the invention the hydrophobic compounds such as plant sterols are incorporated predominantly into the aqueous phase of the fat spread in a largely insoluble form. This embodiment also relates to a process for the preparation of fat spreads with acceptable organoleptic properties which does not necessarily require expensive chemical or mechanical treatment of the above said plant sterols. The process according to this embodiment of the invention is to bring the hydrophobic compound, such as insoluble plant sterols, and milk derived or other coating ingredients into contact in the aqueous phase of the fat spread, preferably at elevated temperature, prior to the emulsification of the above said aqueous phase with a fat phase to produce the desired emulsified product which has a taste and appearance similar to butter and which is spreadable.

15       A further embodiment of the invention also relates to a method for the preparation of a dry ingredient containing hydrophobic compound, such as plant sterol, and milk derived solids which can be used directly in the production of food products or beverages by conventional manufacturing methods.

20       Another aspect of this invention relates to the discovery that the addition of plant sterols to a fermentable raw material does not interfere with lactic acid bacteria fermentation. This invention also relates to food products which are produced by the fermentation of a mixture of raw

material and plant sterols with lactic acid bacteria. The process of the invention brings together a raw material such as milk and conventional milk-derived ingredients and plant sterols, which are virtually insoluble in the fermentation mixture, to produce a range of fermented food products that have similar texture, appearance and flavour to, or are substantially indistinguishable from, the corresponding product compositions which are made with no plant sterols added. The use of other food ingredients such as thickeners, sweeteners, flavourings, colouring agent and fruit products to improve the texture, appearance and flavour of the plant sterol supplemented yoghurt and other products is also disclosed.

This invention brings to the market a range of novel hydrophobic compound-supplemented, such as plant sterol-supplemented, fermented food products which were not previously known.

In accordance with one broad aspect of the present invention there is provided a food product which is produced by the fermentation of a mixture of a fermentable raw material and plant sterols using lactic acid bacteria, and which has a similar texture, appearance and flavour to, or is substantially indistinguishable from, the corresponding product composition made with no plant sterols added.

In accordance with another aspect of this invention, a fermented milk product is provided which is produced by the fermentation of a mixture of milk and/or conventional milk-derived ingredients and plant sterols using lactic acid bacteria, and which has a similar texture,



appearance and flavour to, or is substantially indistinguishable from, the corresponding product composition which is made with no plant sterols added.

In accordance with yet another aspect of this invention, a range of  
5 yoghurts, which are produced by fermentation of a mixture of milk and/or conventional milk-derived ingredients and plant sterols, of different type, composition and fat content as well as yoghurt products are provided where the above said yoghurts and yoghurt products have a similar texture, appearance and flavour to, or are substantially indistinguishable  
10 from, the corresponding product compositions which are made with no plant sterols added.

Yet another aspect of this invention is to provide a process for the preparation of the above said fermented food products, where plant sterols are added to the fermentation mixture prior to inoculation of a fermentable  
15 raw material with micro-organisms.

Yet another aspect of this invention is to provide a process for the preparation of the above said fermented milk products and yoghurts and yoghurt products, where plant sterols are added to the fermentation mixture prior to and/or during the heat treatment step.

20 Still another aspect of this invention is to provide a method for the supplementation of yoghurt and yoghurt products, by their admixture with plant sterols and their derivatives, where the sterols are added either in the pre-coated form or as a fine powder, until a homogenous product is

obtained and where the above said homogenous product has a similar texture, appearance and flavour to, or is substantially indistinguishable from, the corresponding product composition which is made with no plant sterols added.

5

## DETAILED DESCRIPTION OF THE INVENTION

### DEFINITIONS

In the present invention the following terms and definitions are  
10 used:

- In the context of the hydrophobic compounds of the invention, the term "which can be shown to be beneficial to human health" is understood to mean any physiologically active compound which is not a nutrient and can be shown to prevent or reduce the risk or ameliorate the conditions or  
15 symptoms of a disease when taken regularly as a part of a diet.
- The term "hydrophobic compounds" of benefit to human health is understood to mean any organic substances which are substantially insoluble in the aqueous phase of a food product or beverage at or close to the concentration which is thought to be necessary to provide the above  
20 said health benefit.
- The term "emulsified fat products" is understood to mean any products which comprise oil-in-water or water-in-oil emulsion or a combination of the two.

- The term "fat spread" is understood to mean a product containing at least 5% fat, has a consistency and taste similar to that of butter and which is spreadable on a surface of solid foods such as a slice of bread.
- The definitions of "low fat" and "very low fat" when referring to, for  
5 example, a fat spread or a yoghurt are as defined by legislation or relevant regulatory authorities. "Low fat" and "very low fat" spreads may have less than 5% fat.
- The term "food acceptable" is understood to mean that a process  
10 and/or a substance which is permitted for use in the manufacturing of food products and/or beverages or a process and/or a substance for which such a permission can be obtained.
- The term "acceptable organoleptic properties" of a product is  
understood to mean that the product may be organoleptically distinguishable but its overall organoleptic quality is comparable to the  
15 analogous products which are currently on the market. In other words, the organoleptic properties of the modified food product or beverage are generally acceptable to the consumer.
- The term "lactic acid bacteria" is understood to mean micro-organisms which are capable of producing lactic acid in the course of  
20 fermentation.
- The term "fermented food products" is understood to mean any edible products, in a solid or liquid form, which are produced by contacting

a fermentable raw material with a particular micro-organism for the time sufficient for the fermentation to occur.

- The term "fermentable raw material" is understood to mean any material which is or can be fermented by conventional manufacturing  
5 methods.

- The term "fermented milk products" is understood to mean the products which are produced by incubating milk or a fermentable raw material derived therefrom with a particular micro-organism or a mixture of micro-organisms. As used herein "fermented milk products" include  
10 various types of regular yoghurt, low fat yoghurt, non fat yoghurt, kefir, ymer, buttermilk, butterfat, sour cream, sour whipped cream as well as fresh cheeses such as quark and fromage frais, each with its respective modifications.

- The term "milk" is understood to mean conventional milk as well as  
15 equivalent compositions formed by suitable admixtures of butterfat-containing milk products and/or milk solids.

- The term "yoghurt products" is understood to mean foods and beverages containing yoghurt or yoghurt derived ingredients in not insubstantial quantities. As used herein "yoghurt products" include  
20 yoghurt drinks, instant yoghurt compositions, ice creams, frozen desserts and the like.

- The term "substantially indistinguishable products" is understood to mean products which appear the same when inspected by the naked eye

and which cannot be instantly distinguished organoleptically by an assessor.

**PREPARATION OF A COMPOSITION SUITABLE FOR INCLUSION IN A  
5 FOOD PRODUCT OR BEVERAGE WHICH COMPOSITION CONTAINS  
PLANT STEROL, LYCOPENE OR OTHER HYDROPHOBIC  
COMPOUND**

The invention provides a method for making a composition suitable for inclusion in a food product or beverage the method comprising the step  
10 of combining a plant sterol, lycopene or other hydrophobic compound which can be shown to be beneficial for human health with a component which is acceptable as a food additive, wherein the component which is acceptable as a food additive interacts with the surface of the hydrophobic compound. Preferably, any undesirable sensation of powderiness in the  
15 mouth due to the plant sterol, lycopene or hydrophobic compound as defined is masked.

Typically, the hydrophobic compound is any one of a plant sterol (some of which are described in more detail below), lycopene and other carotenoids and poorly water soluble anti-oxidants, or a combination  
20 thereof.

Suitably, the component acceptable as a food additive is any one of a food-acceptable polypeptide, polysaccharide or low molecular weight substances with similar chemical functionality/functional groups.

As is discussed in more detail below, the food-acceptable polypeptide may conveniently be a milk-derived or soya-derived protein.

Any other food-acceptable polypeptides would be suitable provided that they can interact with the hydrophobic surface of sterols, lycopenes or  
5 other hydrophobic food compound beneficial to human health. Specific polysaccharides include, for example, modified cellulose, pectins and starch.

Typically, the hydrophobic compound is one which is solid at room temperature; preferably, the hydrophobic compound is a plant sterol or  
10 lycopene or a combination thereof.

As is described in more detail below, it is preferred that the hydrophobic compound is contacted with the component acceptable as a food additive in aqueous suspension. It is particularly preferred that the composition suitable for inclusion in a food product or beverage is one  
15 which allows the hydrophobic compound, such as plant sterol or lycopene, to be in the aqueous phase once incorporated into the food product or beverage.

Preferably during the preparation of the composition the ratio of hydrophobic compound to coating agent is from about 0.1 to about 0.4  
20 w/w. Preferably the amount of coating agent allows the particles to be fully coated.

The composition may be formed *in situ* in the process of the preparation of a food-product or beverage, or it can be prepared

separately and added during the manufacture of the food product or beverage.

One of the difficulties of incorporating plant sterols, lycopenes and other hydrophobic compounds which can be shown to be beneficial to human health, into predominantly aqueous-based food products and drinks is their extremely poor solubility in water. For example, plant sterols are hydrophobic substances and, as obtained, do not disperse well in water. This behaviour is generally attributed to their poor wettability in aqueous solutions. It was unexpectedly found that plant sterols can be satisfactorily admixed into milk, preferably at elevated temperatures as specified herein and that their addition does not interfere significantly with lactic acid bacteria fermentation. The latter result is especially surprising because sterols are biologically active compounds which are present in many type of cells, for example in cell membranes, and are known to influence metabolic activity and growth of micro-organisms.

It was discovered that the addition of as much as about 5 gram of plant sterols per 100 gram of milk which was fortified with 1.5 gram of high protein whey powder has no appreciable effect on the rate of lactic acid bacteria fermentation as determined, for example, by the time necessary for the resulting yoghurt to reach a pH of about 4.5. Even more surprising, there was little difference in the appearance of the resulting yoghurt and the corresponding conventional product made with no plant sterols added. This is a surprising finding given that the solid content of fortified skimmed

milk used was only about 12 gram of solid per 100 ml.

It was further found that the organoleptic properties of yoghurt are appreciably dependent on whether plant sterols are added to the fermentation mixture before, during or after the milk heating step. This heating step is used by most manufacturers of fermented milk products such as yoghurts, regardless of the variety, to achieve the denaturation of milk proteins and to pasteurise the milk. Thus, when the same plant sterols were added to the milk after cooling but prior to the addition of starter culture containing lactic acid bacteria, the organoleptic properties of the product was poor as determined by tasting. Typically, these products were gritty and left an undesirable sensation in the mouth. However, when plant sterols were added to milk during or prior to the heating step, preferably blended in at the stage of homogenisation of the milk, the resulting yoghurt had a texture, appearance and flavour like the corresponding product composition which is made with no plant sterols added.

It was also found after extensive study and research that a product with even better organoleptic properties i.e. the product which is substantially indistinguishable from the corresponding composition which is made with no plant sterols added, can be obtained when plant sterols are pre-coated with certain food additives, preferably those derived from milk, preferably milk proteins or other proteins, prior to adding the above said coated sterols to the aqueous phase of a food product or a beverage.



The coating can be achieved by stirring a suspension of plant sterols with the coating agent or the coating agent can be added to the suspension as a solid or as a concentrated solution. In addition sterols can be suspended and homogenised in a solution of the coating agent. Although  
5 milk proteins such as caseins and their salts, whey proteins and milk derived solids such as milk powder, whey or buttermilk are preferably used, those skilled in the art will instantly recognise that other food acceptable coating agents such as proteins from sources other than milk, and polysaccharides and even low molecular weight substances with  
10 appropriate functionality may also be employed.

Typically, a suspension or slurry of plant sterols in water containing from about 10% to about 30%, and preferably from about 15% to about 25% of sterol by weight, is prepared by extensive homogenisation using conventional methods and a small volume of a concentrated aqueous  
15 solution of, for example, sodium caseinate or whey protein or milk powder is then added. The above said concentrated solution typically has a solid content from about 8% to about 20% by weight, preferably from about 12% to about 18% by weight. The added volume may vary from about 1.0 to about 10 parts per 10 parts, preferably from about 1.5 to 3 parts per 10  
20 parts of the suspension of sterols.

The suspension of plant sterols can also be prepared by homogenising the said sterols in a pre-prepared solution of, for example, casein, sodium caseinate, whey proteins, whey, milk powder or buttermilk

or even milk itself. It is also surprising that a fine suspension of plant sterols in water can be produced in the absence of surfactants such as, for example, polyoxyethylene sorbitan esters or sodium dodecyl sulphate, and without grinding plant sterol with admixers e.g. sugars as was previously practised in the art (see, for example, US 3,085,939; US 4,195,0840; 5 US3,881,005; GB 934,686). The solid content of the coating agent in the final admixture with plant sterols can vary from about 0.1% to about 2.0% by weight, preferably from about 0.5% to about 1.5% by weight.

It was found that the coating can be accomplished by a variety of treatments. The suspension of sterols can be stirred for some time in the 10 presence of the coating agent at ambient temperatures, or heated and then cooled down or treated by changing pH. For example, the plant sterol suspension prepared by any of the methods described herein, can be left stirring at ambient temperatures, preferably from about 0°C to about 25°C for a period of time from about 20 minutes to about 16 hours, 15 preferably from about 1 hour to about 5 hours. Also, the suspension can be heated to a temperature from about 30°C to about 100°C, preferably from about 40°C to about 60°C, preferably with stirring. The heating time may vary from about 15 minutes to about 2 hours, depending on the final 20 temperature and the rate of heating and cooling. Also, if substances with ionisable functional groups are employed such as proteins or polysaccharides, the pH of the suspension can be altered by adding a dilute acid or base such as to neutralise substantially the net charge in the

molecule of the coating agent, preferably to a pH which is near the isoelectric point of the coating agents present in the suspension. In the case of sodium caseinate, the pH is typically adjusted with a dilute acid to a pH of between about 4.0 and about 5.0, preferably to pH 4.5 to

5 protonate the available carboxyl groups, and the slurry is left stirring for a period of time from about 1 hour to about 16 hours, after which time the pH is adjusted back to the preferred values of 6.1-6.7. Alternatively the coating can be achieved by precipitating plant sterols into solution containing a coating agent. For example, sterols can be dissolved in an

10 organic solvent, preferably a water-miscible organic solvent, and the resulting solution is then poured into water containing milk-derived solids or proteins or other suitable coating agents as described herein. All these methods were found to be satisfactory. The coated plant sterols can also be recovered in a dry form using conventional drying techniques,

15 preferably spray drying.

It was also found that coating according to the present invention can be successfully obtained when plant sterols are combined with other non-sterol food additives. Such additives can be physiologically active food substances known to those skilled in the art or other substances

20 added to enhance, for example, the bioavailability of plant sterol. For example, prior to the coating as described herein, plant sterols can be co-precipitated with lycopenes which are also known to reduce plasma level cholesterol in humans, but by the mechanism different to that of plant

sterols, or lecithin. The additives can be used in the amount as small as less than 0.1 part to 1 part of plant sterols (e.g. lycopene) or as high as 0.95 part to 1 part of plant sterols (e.g. lecithin).

The treatments described herein usually result in the coating of  
5 plant sterol particles in the suspension by proteins and/or other components present in, for example, milk powder. Consequently, the coated plant sterols display properties which are different from that of uncoated material. It was observed, for example, that spray dried sodium caseinate coated plant sterols can be suspended in water with relative  
10 ease and on stirring form a suspension which is rather similar in appearance to milk. It was found that the separation of plant sterol particles in this suspension occurs significantly more slowly than in analogous suspensions made from uncoated sterols.

The formation of the "coat" on the surface of sterol particles can be  
15 directly observed by confocal laser microscopy when proteins, for example, are labelled with an appropriate fluorescent dye under conditions similar to those described by Aherne *et al* (*J. Am. Chem. Soc.* (1996) **118**, 8771-8772). Both the labelling methods and the technique of fluorescent microscopy are well known in the art.

20 The common feature of the treatments described herein is that the coating agent is encouraged to adhere to the surface of the plant sterol particles. In the case of proteins, this can be achieved, for example, by substantial neutralisation of the net charge of the protein by adjusting the

pH to values close to the protein's isoelectric point or by any other treatment leading to their partial denaturation, e.g. heating or presence of organic solvents, or simply by prolonged incubation at ambient temperatures. Thus, in the case of whey proteins which are known to denature at relatively low temperature, mild heating is the preferred treatment. These examples of treatments are not exhaustive and those skilled in the art will instantly recognise that numerous other treatments and modifications to the coating procedures disclosed herein can be used to exercise this aspect of the present invention. It is asserted that in addition to prolonged incubations, pH adjustment and heat treatment, any other treatment or conditions which may be reasonably expected to promote the interaction between the coating agent and the surface of the plant sterol particles can be fruitfully employed. A desirable property of the resulting coated composition is an ability to easily form an aqueous suspension.

Ideally, the coating composition should expose functionality. This means that the coating component which is acceptable as a food additive, which when it interacts with the surface of the plant sterol, lycopene or other hydrophobic compound which can be shown to be beneficial to human health, also has a functionality which when exposed to an aqueous phase encourages the coated particles to form a suspension in the aqueous phase.

Generally among the coating agents, those which have ionisable groups, preferably carboxyl groups, with a pKa between about 3.5 and about 7.5 and/or those whose solubility in water depends strongly on pH and/or temperature of the aqueous solution or suspension are preferred.

5 Also, the coating agents must be approved for food use and be compatible with the process for the preparation of food products and beverages according to the present invention. Those skilled in the art will recognise that coating agents others than milk derived proteins such as other proteins and various functionalised polysaccharides as well as  
10 appropriately functionalised low molecular weight substances, may also be used. Appropriately functionalised low molecular weight substances preferably contain at least one carboxyl and at least one hydroxyl group.

It was further found that the coating can be accomplished *in situ* i.e. directly in milk, preferably in milk fortified with milk derived solids, as well  
15 as by using the separate coating procedures described herein. The incubation of plant sterols with milk or reconstituted milk or milk fortified with whey proteins or any other suitable additives prior to or after homogenisation of milk, preferably at temperatures between about 40°C and 95°C is sufficient to achieve the coating which according to the  
20 present invention gives acceptable appearance and organoleptic properties of the final product.

Although not wishing to advance or be bound by any particular theory, it is assumed that sterol particles coated with milk proteins, interact

during or after the heating stage with casein micelles and whey proteins and/or other milk components in solution and that these interactions are important for integrating the coated plant sterols into the product such as, for example, yoghurt. It is assumed that it is these interactions that lead to the production of yoghurts which have a similar texture, appearance and flavour to, or are substantially indistinguishable from, the corresponding product compositions which are made with no plant sterols added. Those skilled in the art will instantly recognise that the coated plant sterols according to this invention may also be used in a variety of other foods and food products as is described in more detail below, particularly in the Examples.

Further details of the method of the invention are given below in relation to fermented dairy products and emulsified fat products.

It will be appreciated that the invention also provides a composition suitable for inclusion in a food product or beverage obtainable by the method for making a composition suitable for inclusion in a food product or beverage the method comprising the step of combining a plant sterol, lycopene or other hydrophobic compound which can be shown to be beneficial for human health with a component which is acceptable as a food additive, wherein the component which is acceptable as a food additive interacts with the surface of the hydrophobic compound.

Similarly, the invention provides a composition suitable for inclusion in a food product or beverage comprising a plant sterol, lycopene or other

hydrophobic compound which can be shown to be beneficial for human health with a component which is acceptable as a food additive coated on the surface thereof.

A composition of the invention which is useful in food and pharmaceutical formulations, and one which is particularly useful in relation to emulsified fat products, is a dry ingredient which is obtained by drying, preferably spray drying, of a mixture containing a milk derived solid and plant sterols, where the milk derived solid and the plant sterols are contacted in an aqueous suspension, and where the milk derived solid is selected from the group consisting of casein, caseinate, whey protein, whey, milk powder, buttermilk and butterfat, and where the plant sterols are selected from the group comprising  $\beta$ -sitosterol,  $\beta$ -campesterol,  $\beta$ -stigmasterol,  $\beta$ -sitostanol,  $\beta$ -campestanol and  $\beta$ -stigmastanol and their carboxylic acid esters, and a mixture thereof.

Typically, the milk-derived solid and the plant sterol are contacted at a temperature above 40°C, preferably with stirring. Preferably the contact is carried out at a temperature and exposure time combination which is sufficient for pasteurisation.

Furthermore, in addition to plant sterols, the present invention can also be practised with other food additives which are known to be beneficial to human health, for example lycopene, provided that these additives are sufficiently hydrophobic to enable their coating according to the present invention, preferably with proteins or milk-derived solids or any



other coating agent according to the method disclosed herein. The above said additives can be coated separately and used in foods and beverages on their own or premixed, or co-precipitated with plant sterols as exemplified herein.

5

## PRODUCT PREPARATION

The methods and products detailed above are useful in the preparation of a variety of food products and beverages. Thus, the invention provides a method for preparing a food product or beverage  
10 which is supplemented with a plant sterol, lycopene or other hydrophobic compound which can be shown to be beneficial for human health the method comprising the step of (1) carrying out the method for making a composition suitable for inclusion in a food product or beverage the method comprising the step of combining a plant sterol, lycopene or other  
15 hydrophobic compound which can be shown to be beneficial for human health with a component which is acceptable as a food additive, wherein the component which is acceptable as a food additive interacts with the surface of the hydrophobic compound *in situ* during the preparation process or (2) adding at an appropriate stage during the preparation  
20 process a composition obtainable by the method of step (1) above, or a composition suitable for inclusion in a food product or beverage comprising a hydrophobic compound which can be shown to be beneficial for human health with a component which is acceptable as a food additive

coated on the surface thereof, or (3) a combination of (1) and (2).

The appropriate stage in a manufacturing process for adding the composition (eg coated plant sterol) is a stage where the food additive  
5 comprising the "coat" material can be added without substantially altering the overall manufacturing process or the properties of the final product. For example, in the case of emulsified fat spreads this is a stage of formulating the aqueous phase of the product prior to emulsification, whilst in the case of manufacturing a yoghurt this is the stage of fortifying milk  
10 with additional whey solids and in the case of the preparation of yoghurt fruits this is the stage of heating up the initial pre-mix prior to the addition of the fruit.

Typically, the hydrophobic compound resides in the aqueous phase of the final food product or beverage. For example, preferably it resides in  
15 the aqueous phase of an emulsified fat product.

The invention also provides a food product or beverage which is supplemented with a hydrophobic compound which can be shown to be beneficial for human health obtainable by the method mentioned immediately above.

20 Preferably, the hydrophobic compound is a plant sterol or lycopene or combination thereof. Preferably, the component which is acceptable as a food additive is a milk-derived solid.

The following is a description of certain food products and

beverages which have been supplemented with the hydrophobic compound which can be shown to be beneficial to human health. Some of the processes do not make use of coated hydrophobic compounds but, for the avoidance of doubt, processes, food products and beverages made  
5 using such processes are part of the invention described herein.

### **Fermented dairy products**

An extensive study and research was carried out on the processing methods and conditions that are required to produce products which are fermented with lactic acid bacteria, preferably fermented milk products,  
10 preferably yoghurt and yoghurt products, with acceptable organoleptic properties, where the above said fermented products are supplemented with plant sterols, preferably with the plant sterols coated according to the present invention.

In general, the process by which the fermented milk products of the  
15 present invention are made involves forming a milk base by admixing, preferably at an elevated temperature of about 30°C or above, a fermentable dairy product including whole milk, low fat milk, skimmed milk, skim milk, condensed milk, evaporated milk, milk powder including non-fat dry milk powder (NFDM), buttermilk, cream and the like, and optionally  
20 thickeners and stabilisers, and optionally a sweetener, and optionally adding other sweeteners, flavourings and colorants, and optionally adding vitamins and minerals to the admixture in sufficient quantities to provide a product which is either similar in texture, appearance and flavour to, or is

substantially indistinguishable from, the corresponding product composition which is made with no plant sterols added. The resulting mixture is optionally homogenised by conventional homogenisation means and pasteurised and further heat labile ingredients such as vitamin C are  
5 optionally added. The mixture is inoculated with a suitable micro-organism or a mixture of micro-organisms which produce lactic acid during the fermentation and which are suitably chosen for the kind of product to be made, and the inoculated mixture is maintained under a carefully controlled temperature regime until the desired flavour, texture and  
10 appearance are achieved.

In making the fermented milk products of this invention, it is preferred to use cows milk as defined herein or ingredients derived therefrom. However other milk, for example, ewe milk, goat milk, buffalo milk and others as well as ingredients derived therefrom such as whey,  
15 whey proteins, caseins, caseinates, milk powder, cream, buttermilk or butterfat, may also be used. It was found that regardless of whether yoghurts are made from fresh milk or reconstituted milk and whether the milk and milk powder used were skimmed, semi-skimmed or whole, the yoghurt which is supplemented with plant sterol according to the present  
20 invention has a similar texture, appearance and flavour to, and often is substantially indistinguishable from, the corresponding product composition which is prepared by the same method using the same ingredients but with no plant sterol added. Furthermore, it was found that

a substantial part of the milk or milk powder, preferably between about 20% to about 70% on a dry weight basis, can be replaced by soymilk. Also, dry milk and milk powder which contain the coated plant sterols in accordance with the present invention, may be utilised as one of the  
5 ingredients.

It was further found that yoghurts which have similar texture, appearance and flavour to, or are substantially indistinguishable from, the yoghurt of the same composition but with no plant sterol added, can be prepared using a variety of plant sterol preparations from different  
10 suppliers (Sigma Aldrich Chemical Co (Dorset, UK), Fluka Chemicals (Dorset, UK) and ICN Biomedicals Inc (Ohio, USA)). The plant sterols preparations may contain as little as about 50% or as much as about 95% of a single compound such as  $\beta$ -sitosterol and different quantities of other plant sterols such as campesterol, stigmasterol and the like. 5 $\alpha$ -saturated  
15 derivatives of the above said sterols, for example,  $\beta$ -stanol can also be used. It was further found that some technical grade sterol preparations, typically containing about 50% of plant sterol or less desirable for use in the present invention in the form as obtained from the supplier. These preparations have a distinct and unpleasant smell and must be further  
20 purified to remove the odorous impurity. This can be accomplished by conventional methods such as chromatography or precipitation from organic solvents or an organic solvent-water mixtures, preferably with organic solvents that are permitted in food processing.

It was also found that, in general, yoghurts which are supplemented with sitosterol have somewhat better organoleptic properties than those prepared with stanol, both at the same loading. It was also found that esterification of the free hydroxyl group with a carboxylic acid in the molecule of stanol results in some improvement in the organoleptic properties of the product (this esterification can be accomplished by numerous methods well known in the art) and the yoghurt which has a similar texture, appearance and flavour can be produced using the above said esters. It will therefore be obvious to those skilled in the art that plant sterol preparations with different content of individual components as well as other derivatives of plant sterols may also be used according to the present invention. For example, plant sterols which are obtainable from rice bran oil such as cycloartenol and 24-methyl-cycloartenol, and from sheanut such as amyirin, lupeol and butyrospermol may also be used.

It was also found that yoghurts with somewhat better organoleptic properties are obtained when plant sterols are used as a fine powder rather than in the form of larger particles. In the latter case, it is preferred to break the particles by high power homogenisation or any other form of shearing to improve the organoleptic properties of plant-sterol supplemented fermented products such as yoghurt and the like.

It is preferable that the yoghurt according to this invention contains milk solids in quantity from between about 10% to about 25% by weight. Satisfactory results are also obtained when the milk solids content of the

yoghurt prior to pasteurisation is about 15%, for example, between 12% and 21%. The butterfat content may be between less than 0.2-0.5% for non fat or very low fat yoghurt to about 10% for a Greek style yoghurt. In order to prepare a low fat or non fat yoghurt or yoghurt product, the use of fat contributing ingredients must be carefully controlled. In as much as the primary fat contributing ingredient in yoghurt is generally milk, a low fat content may be achieved readily by selecting a skimmed milk with a butterfat content of less than about 0.4% and preferably less than about 0.2% by weight. Typically, for the production of yoghurt use is made of starters consisting of *Lactobacillus bulgaricus* and *Streptococcus thermophilus*.

Typically the yoghurts of this invention are made by making the fermentation mixture by blending the desired amount of milk, to which whey proteins in quantity from about 1.0 gram to about 3.0 gram per 100 gram of milk, preferably about 1.5 gram per 100 gram of milk, with the desired amount of plant sterols, typically from about 1.5 gram to about 5 gram per 100 gram of milk. The pH of the mixture is adjusted to the preferred value of 6.1-6.7, if necessary, by adding an acidifying agent. The acidifying agent may be a food quality acid, for example, lactic or citric acid. In a commercial scale process it is preferred to employ a supplemental size reduction step after the slurry preparation but prior to admixture with the whey containing milk (yoghurt base). Suitable supplementary size reduction techniques include, for example, passing the

slurry through a colloidal mill, or a two stage homogeniser followed by passing the slurry through mesh screeners, and using a high speed shear impeller blender in which the slurry is prepared. The slurry can be passed through the homogeniser for one, two or more passes sufficient to provide  
5 good suspension. Homogenisation prior to pasteurisation is optionally carried out in order to promote a smooth texture in the yoghurt and aid in the dispersion. Any prior art homogenisation conditions can be satisfactorily employed in the present invention.

The ingredients of the fermentation mixture are then treated at high  
10 temperature, preferably at legally specified time and temperature conditions, using techniques and equipment well known in the art. While the heat treatment may be carried out at many different temperature and exposure time combinations, the body and texture of the yoghurt is generally improved by the application of heat treatment regimes which are  
15 known from the art to be sufficient to cause a substantial denaturation of whey proteins. Also it is preferred to carry out the heat treatment under such a combination of temperature and exposure time that is sufficient to pasteurise the fermentation mixture i.e. to inactivate or deter the development of undesired micro-organisms. Typically the heat treatment  
20 is performed at a temperature in the range of 60°C to about 110°C, preferably at about 80°C to 95°C, and optionally under high pressure, and optionally, with stirring. Care should be exercised not to heat the mixture longer than necessary as deleterious effects can result. It was generally



found that regardless of specific heating regimes, the yoghurts which are supplemented with plant sterols according to the present invention have a similar texture, appearance and flavour to, or are substantially indistinguishable from, the corresponding product compositions which are prepared by the same method from the same ingredients but with no plant sterol added.

The heat treated and pasteurised mixture is cooled in one or more stages, optionally with stirring, to a temperature in the range of about 37°C to about 50°C, preferably to between about 40°C to about 45°C and it is inoculated with a micro-organism to initiate the fermentation. The micro-organism used in the fermentation is essentially a lactic acid bacterium which is preferably used in a mixture with other yoghurt producing micro-organisms. The micro-organism can be one of several yoghurt producing cultures. It is preferred to use *Lactobacillus bulgaricus*, a lactic acid forming rod-shaped bacterium, and a *Streptococcus thermophilus*, a coccoid bacterium, either alone or together with other suitable yoghurt producing micro-organisms. These micro-organisms can be used in equal amounts or their relative quantities can be adjusted as desired, in accordance to the preference of the yoghurt producer. *Lactobacillus acidophilus*, or other yoghurt producing micro-organisms can be added to the *Lactobacillus bulgaricus* - *Streptococcus thermophilus* combination above or in other combinations. Other examples of species of *Lactobacillus* and *Streptococcus* which may be used to practice the

present invention include *Lactobacillus bulgaricus*, *helveticus*, *jugurti*,  
*lactis* or *acidophilus* and *Streptococcus thermophilus*, *ceморris*, *lactis* or  
*diacetilatis*. *Bifidobacterium* sp., for example, *Bifidobacterium breve*,  
*bifidum*, *infantis*, *longum*, *adolescentis* and *Lactococcus* sp., for example,  
5 *Lactococcus casei*, can also be optionally added. These micro-organisms  
can be obtained as starter cultures from commercial sources such as, for  
example, Chr. Hansen A/S of Denmark or Gist Brocades NV of the  
Netherlands or from public culture collections, or isolated from natural  
sources. Species and strains which are selected or genetically modified  
10 for higher resistance to phage infection or to provide yoghurts with  
superior properties or under superior manufacturing may also be  
employed.

It was generally found that regardless of specific micro-organisms  
used and the source of the starter culture, the yoghurts that are  
15 supplemented with plant sterol according to the present invention have a  
similar texture, appearance and flavour to, or are substantially  
indistinguishable from, the products prepared by the same method from  
the same ingredients with the same micro-organisms but with no plant  
sterol added.

20 Those skilled in the art will instantly recognise that fermented milk  
products, including yoghurts, can be prepared with the addition of more  
specialised cultures such as the above mentioned Bifidus bacteria to  
obtain products with health benefits in addition to those provided by plant

sterol. It is assumed to be obvious that such cultures as well as other food additives of proven benefit to human health can be added to the fermented milk products such as yoghurt, in addition to plant sterols, provided that the above said additives do not interfere with sterols  
5 chemically and do not interfere with the process of making yoghurts and other fermented milk products.

The fermentation is continued for an appropriate time until the yoghurt composition reaches a pH in the range of about 3.8 to about 5.0, preferably from about 4.0 to about 4.7, a pH about 4.3-4.6 being preferred.  
10 At this time the consistency of the yoghurt is denominated as firm as is understood in the yoghurt art. Appropriate time for fermentation is generally in the range of about 3 hours to about 20 hours, preferably from about 4 hours to about 6 hours. Longer and shorter times may be used but generally are unnecessary. Fermentation is carried out in the  
15 fermentation vessel or in yoghurt containers.

It was also found that at a higher loading of plant sterols, typically in excess of about 3.0 gram per 100 gram of milk, a certain amount of precipitate may sometimes be formed at the bottom of the container. It was further found that this precipitation can be reduced considerably or  
20 avoided altogether by, for example, increasing the initial amount of micro-organisms used in fermentation or by increasing the fermentation temperature or by the combination of the above or by any other method which reduces the fermentation time. In general a lower fermentation time

is preferred for the preparation of set type yoghurts with higher sterol loading as specified herein. It was also found that the formation of precipitate can be substantially reduced or even avoided altogether by using a continuous or semi-continuous fermentation techniques which are well known to those skilled in the art, with the use of pre-fermenting and coagulation tanks being preferred. It was further found that the quantity of precipitate can be reduced or avoided and the organoleptic properties of the product improved by the use of an appropriate stabilising or thickening agents.

10       According to the yoghurt art, a stabilising or thickening agent can be optionally added in a suitable quantity to retard any whey separation during storage and transportation (the process known as syneresis). These agents are also known in the art to offset the influence of added ingredients such as fruits or flavouring agents. It is preferred to use stabilising and/or thickening agents with low fat and non fat yoghurts that are supplemented with plant sterols in accordance with this invention. It is also preferred to use stabilising or thickening agents with yoghurts according to the present invention which are supplemented with higher quantities of plant sterol from about 2.0 gram to about 5.0 gram by weight per 100 gram of the final product, regardless of the fat content. It was found that the use of the above said stabilising and thickening agents improves the texture and the mouthfeel of the yoghurts, often producing a yoghurt which is substantially indistinguishable from the composition which

is prepared by the same method from the same ingredients but with no plant sterol added.

Conventional stabilising and thickening agents can be used in the normally required quantity to retain the desired consistency and texture of the yoghurt. A thickening agent is preferably water soluble and one that is approved for use in fermented food products such as yoghurt. Generally the amount of the thickening agent used does not exceed about 0.5% to about 1.0%, depending on yoghurt composition, the other additives used and, in particular, the type of thickening or stabilising agent employed.

Such stabilising or thickening agents include plant exudates (e.g. gum arabic), seaweed extracts (e.g. alginates), plant and seed gums (e.g. guar gum), plant extracts (e.g. pectin) and animal derived products (e.g. gelatin). These commercial thickeners and stabilisers are soluble in water and milk and therefore can be incorporated into the yoghurt mix prior to the fermentation process or added at a later stage in the preparation of conventional stirred yoghurt. Also, other polysaccharides such as xanthan gum, locust gum and carrageenan and pectin derivatives such as partially demethylated pectins, may be used either on their own or in combination with emulsifiers such as mono/diglycerides, lecithin or polysorbates.

It was further found that the even when the above mentioned precipitate was formed after fermentation, it can be blended into the yoghurt by conventional stirring to produce stirred yoghurts. Once stirred in, no further precipitation was observed during a storage period of several

weeks. However, some powderiness was still noted in the organoleptic assessment of the stirred yoghurts when prepared at loadings in excess of about 2.5 gram of sterol per 100 gram of milk and especially at levels exceeding 3 gram per 100 gram of the final product. It was then  
5 discovered that this sensation of powderiness can be greatly reduced and in most cases eliminated completely by the additional of a fruit and/or a flavouring which are normally used in conventional yoghurt varieties. It is sometimes advantageous to add to the yoghurt an amount of thickening agent to offset the influence of such added fruits or flavouring agents. The  
10 thickening agents as described herein can be used in the normally required amount to retain the desired texture and consistency and to improve the organoleptic properties of the final yoghurt.

The fruit is added in the conventional manner either as a dispersion in the pasteurised fermentation mix or it can be added aseptically in the  
15 form of a pasteurised fruit preparation. The fruit can be conventionally added to the bottom of the container in which yoghurt is sold before adding the yoghurt or it can be packed in a separate compartment to be mixed with yoghurt prior to consumption. The yoghurt container, with the fruit preparation at the bottom can, after capping, be inverted to cause the  
20 fruit preparation to come to the top of the yoghurt surface. The fruit preparation can also be mixed into the yoghurt if stirred or Swiss style yoghurt is desired.

The amount of the fruit added can vary from about 5% to about 40%, preferably from about 10% to about 35% by weight of yoghurt. The fruit component may optionally contain, in addition to conventional sugars, monosaccharide sugars having a high sweetening effect. For example, it is well known in the art that the syrup for the fruit pieces can be advantageously prepared using a combination of fructose and sucrose as the sweetener. Generally, the following compositions of, for example, strawberry fruit products: Swiss style fruit product comprising 40-55% strawberries, 30-40% sugar, 1-3% modified starch and Sundae style fruit product comprising 35-45% strawberries, 15-25% sugar, 15-20% corn syrup, 2-4% modified starch, both with optional flavouring and colouring agents, acidifiers and preservatives, are suitable.

The flavouring agent can be optionally added to the pasteurised fermentation mixture. The flavouring agents may be natural or artificial, and it is preferable to use those which impart a fruit flavour such as strawberry, blueberry, peach, cherry, lemon, mango, banana and the like. Other flavours such as vanilla, chocolate and others which are customarily used in the yoghurt art can all be used. The amount of flavouring agent used may vary widely according to the taste, but generally an amount from about 0.1% to about 5.0%, typically from about 0.3% to about 3.0% and preferably from about 0.5% to about 1% by weight based upon the total composition on a dry weight basis, is used.

It may also be desirable or even required when a flavouring agent is employed, to use a sweetening agent to, for example, offset the taste of added compounds. Conventional sweetening agents can be used for this purpose, such as sucrose, fructose, and synthetic agents such as aspartame, saccharin and the like. Other sweet substances, for example, palatinose, palatinite, maltose, maltitol, mesoerythritol, starch syrup, reducing starch syrup and trehalose may also be employed.

Colouring agents may also be used in the preparation of the yoghurt of this invention. A colouring agent can be any acceptable colouring ingredient, preferably appropriate to the flavouring agent. For example, it is preferred to use an appropriate approved red colouring in the case of strawberry flavour and an appropriate approved yellow colouring with banana flavouring. The amount of colouring agent may vary widely but an amount generally from about 0.001% to about 1.0%, typically from about 0.02% to about 0.5% and preferably from about 0.05% to about 0.2% by weight of the total yoghurt composition may be used based on dry weight of the yoghurt composition.

Various yoghurt products can be produced from the yoghurt according to the present invention. For example, a yoghurt drink can be prepared using standard methods well known in the art. Typically, the yoghurt according to this invention is cooled down and water, a food grade acid to adjust pH, stabilisers and flavourings are added and the mixture is homogenised, preferably at elevated temperature, preferably under



pressure. It was generally found that yoghurt products such as yoghurt drinks which are supplemented with plant sterols according to the present invention have a similar texture, appearance and flavour to, or are substantially indistinguishable from, the corresponding product  
5 compositions which are prepared with no plant sterol added.

It was also found after extensive study that plant sterols can be added to fruit yoghurts in a powdered form without significantly impairing the taste of the product when, for example, the said sterols are incorporated into the fruit. It is preferred to use the plant sterols which are  
10 coated in accordance with the present invention, preferably plants sterol coated with milk proteins, and recovered by drying, preferably by spray drying. It was found, for example, that plant sterols, coated with sodium caseins and recovered by drying, when added to water form a fine suspension on stirring at elevated temperature. In appearance this  
15 suspension resembles milk. It was further found that this suspension can be used in place of water for the preparation of fruit products as exemplified in the present invention. For a large scale production it is desirable to subject the above said suspension of plant sterols to a particle size reduction procedure carried out by methods described herein or other  
20 methods well known in the art.

It was also found that even non-coated plant sterols in a powdered form may be used for the production of yoghurt fruit, if ground and sieved to a particle size of about 50 micron, and preferably below about 30

micron, to give a yoghurt with acceptable organoleptic properties. For example, a yoghurt fruit flavouring can be prepared by adding the plant sterols, preferably in a mixture with other solids such as sugar, to a blended fruit at elevated temperature between about 60°C to about 95°C, under conditions of vigorous agitation.

It was further found that fruited yoghurts can be prepared by incorporating plant sterols both into the yoghurt and the yoghurt fruit, to provide products which have a similar texture, appearance and flavour to, or are substantially indistinguishable from, the corresponding product compositions which are made with no plant sterols added. This is an important finding because it enables the production of yoghurts which have a significantly higher loading of plant sterols and acceptable organoleptic properties. Preferably, the plant sterols are unesterified (free) plant sterols: when the plant sterols are added to, or are present in, the yoghurt fruit they may be free or esterified plant sterols; it is preferred that they are free plant sterols.

The equipment used in carrying out the present invention is available commercially and can be conventionally used by those skilled in the art. The equipment utilised must permit the preparation of plant sterol suspensions and slurries, pH adjustments, stirring, homogenisation, controlled heating and cooling, drying and spray-drying, carrying out a fermentation process without contamination and with suitable control of

fermentation temperatures, breaking yoghurt gels, preparing and admixing additional ingredients and final packaging of yoghurts.

Although the present invention is illustrated mainly by the production of yoghurt, those skilled in the art will appreciate that it can be practised successfully in connection with other fermented products and, especially fermented milk products such sour cream, kefir, ymer, buttermilk and the like as well as fresh cheeses, all with their respective modifications. Also, yoghurt products such as instant yoghurt compositions, frozen yoghurt, frozen desserts and the like as defined , herein can be produced from the yoghurt in accordance with the present invention.

#### **Emulsified fat products.**

15        This embodiment of the invention relates to the discovery that when plant sterols are combined with milk derived solids in a suspension to obtain the coated sterols according to this invention, this suspension can be used as an aqueous phase for the manufacture of emulsified fat spreads and, preferably low and very low fat spreads with acceptable organoleptic properties. This is a surprising finding because it is well known in the art that organoleptic properties of emulsified low fat spreads are very sensitive to the composition of the aqueous phase.

Although not wishing to be bound by or advance any particular theory, it is assumed that milk derived solids interact with hydrophobic surface of plant sterol particles and this interaction is sufficient to mask the undesirable sensation in the mouth as well as to enable one to use  
5 conventional manufacturing methods for the incorporation of plant sterols in emulsified fat spreads, preferably low fat spreads. Furthermore when the contact between plant sterol particles and milk derived solids in an aqueous suspension is made according to this invention, the water can be evaporated and the recovered solid can be used directly in the production  
10 of emulsified fat spreads, preferably by adding the said solid to the aqueous phase of the spread prior to emulsification.

According to the present invention, the process for the production of the above said emulsified fat spread involves contacting plant sterols and milk derived solids in an aqueous suspension, preferably at elevated  
15 temperatures, preferably above 40°C. Higher temperatures can also be used and, in this case it is preferred to use the combination of temperature and time exposure which is sufficient to pasteurise the aqueous phase. It is preferred to use milk solids containing a relatively high proportion of proteins such as, for example, sodium caseinate and high protein whey  
20 powder but other milk derived solids such as, for example, milk powder and buttermilk may all be satisfactorily employed. The aqueous phase may contain additional ingredients such as stabilisers, flavouring agents, salts, food grade acids such as lactic and citric acid and preservatives to

increase the shelf life of the product and reduce the risk of microbial contamination.

It was also unexpectedly found that when plant sterols are added to the aqueous phase of fat spreads in the absence of milk derived solids, the emulsification sometimes cannot be carried out satisfactorily, with the emulsion turning into a soft gel. This often happens when emulsification is carried out at too high a temperature and more so when lecithin or polyglycerol esters are used as emulsifiers. The addition of milk derived solids offset this undesirable change in the structure of the emulsion. However, it was also found that the order of addition of milk derived solids and other ingredients such as, for example, gelatine is important. Thus, no gelling was observed on emulsification when plant sterols were contacted with milk solids in the aqueous phase of a fat spread prior to the addition of gelatine or even when plant sterols were contacted with a mixture of gelatine and whey powder for a reasonable time. However, when whey powder is added to plant sterols suspension in gelatine immediately prior to emulsification, the gelling often occurs. Once again, without wishing to be bound by or advance any particular theory it is assumed that the interaction between milk derived solids and plant sterol particles is responsible for this effect.

An extensive study and research was done to elucidate the above said phenomenon and it was found that the addition of lecithin (E322) and/or polyglycerol esters (e.g. E476) to a vegetable oil, even in quantities

as low as 1% by weight or below, increases substantially the solubility of plant sterols in the said oil. Thus, for example the addition of 1% w/w of soy lecithin to sunflower oil increases the solubility of  $\beta$ -sitosterol to about 7% at 60°C, whilst in the absence of lecithin the solubility of  $\beta$ -sitosterol is

5 known to be significantly lower. A similar effect is observed with  $\beta$ -stanol. The addition of polyglycerol esters to oils also increases the solubility of  $\beta$ -sitosterol substantially but glycerol esters (monoglycerides, E471) are relatively inefficient. It was further found that the above said plant sterols solution rapidly turned into a gel on cooling or when a small amount of

10 water is added. A similar observation was made by Ritter *et al* (WO 97/42830) who disclosed the formation of organogels on mixing two sterols of plant origin. However according to the invention of Ritter *et al* at least two different sterols, one of which is esterified, are necessary to achieve the formation of organogels. The gels produced according to the

15 present invention by dissolving plant sterols in an oil matrix containing lecithin and (poly)glycerol esters or the like can be fruitfully exploited for the preparation of cosmetic and food products such as spreads, squeezable margarines and dressing using manufacturing methods that are well established in the art. It was found however that no formation of

20 the gel occurred in the process of the preparation of fat spread, even when a substantially higher amount of lecithin was present in the aqueous phase prior to emulsification, if the mixture of plant sterols and lecithin was coated according to this invention.

When producing low and very low fat spreads according to this invention it is preferred to use stabilisers. The stabilisers which can be used to practice the present invention include gelatine, maltodextrins, starch and modified starch, cellulose and its derivatives and pectins and  
5 their derivatives and other polysaccharides of plant or seaweed origin. However, this is not an exhaustive list and those skilled in the art will instantly recognise that other stabilisers as well as various mixtures thereof can also be successfully employed.

The fat spreads according to this invention can be made with a  
10 different fat content, preferably with fat content below 40%. Conventional fat phases containing vegetable oils such as, for example, sunflower oil, soybean oil, rapeseed oil and the like can all be used as obtained or after hardening or any other chemical or physical treatment as known and acceptable in the art. Animal fats, preferably butter fat, may also be used.  
15 Other ingredients can be optionally added to the fat phase. Examples of the above said ingredients include flavouring and colouring agents and vitamins, preferably those which are conventionally used in the manufacture of fat spreads.

Conventional emulsifiers, preferably mono/di-glycerides (E471),  
20 lecithin (E322) and polyglycerol esters (E476), can be used to produce the fat spreads according to this invention, preferably using the process of this invention. The above said emulsifiers can be used on their own or as a mixture or any combination thereof or in conjunction with other suitable

emulsifiers which are known in the art. The emulsifier can be added to the oil phase or to a part of the oil phase or to the aqueous phase, preferably after contacting plant sterols with milk derived solids.

The spreads according to this invention can be prepared to contain  
5 vitamins, such as vitamins A and D, which are conventionally included into fat spreads or are required to be added by legislation. In addition the spreads according to this invention can be further fortified with additives which are known to be beneficial to human health and preferably those which enhance the desired effect of plant sterols. For example,  
10 antioxidants such  $\alpha$ -tocopherol and/or ascorbic acid can be included into the fat phase or aqueous phase as appropriate. The fat phase can be further supplemented with plant sterols and their derivatives to, for example, increase the overall content of the active ingredient in the final product. Thus, stanol and sterol esters can be included, if desired.  
15 Similarly water soluble additives such as, for example vitamins or minerals, can be included into the aqueous phase, preferably prior to contacting plant sterols with milk derived solids according to this invention.

#### **Other food products and beverages**

20 Those skilled in the art would instantly recognise that the hydrophobic compound-containing composition (such as the sterol-containing additive) described herein can be used in a large number of other food products and beverages such as, for example, alcoholic and



non-alcoholic drinks, soups, sauces, dips, salad dressings, mayonnaise, non-fat spreads, confectionery, bread, cakes, biscuits, breakfast cereals, and non-fermented dairy and non-dairy products.

## 5 Additional supplementation

The food products and beverages according to this invention can also be additionally supplemented with other physiologically active compounds of known benefit to human health. The above said physiologically active compounds do not have to be coated or modified in  
10 any other way, if they are sufficiently soluble in the food product or beverage of the present invention. For example, for low fat products and drinks it is preferred to use those compounds which are soluble in water e.g. soya isoflavones, which are known in the prior art to deter the development of certain cancer and positively influence human health in  
15 many other respects.

## EXAMPLES

### Example 1.

A standard yoghurt base was prepared by adding 1.5% by weight of  
20 high protein whey powder to pasteurised skimmed milk. The mixture was homogenised, heated to 80°C and held at this temperature for 30 minutes. The mixture was then cooled to 45°C and finely divided pasteurised powder of plant sterols (approximately 60% sitosterol and 40%

campesterol and stigmasterol) in the amount of 0, 1.7, 3.5 and 5.1 gram per 100 gram of yoghurt mix was added under aseptic conditions. After admixing plant sterol, the yoghurt mix was inoculated with starter culture containing 1:1 *Lactobacillus bulgaricus* and *Streptococcus*  
5 *thermophilis* (0.5-0.8% w/v), distributed into containers and incubated at 43°C until pH 4.5 was reached. Typically, the fermentation took about 5 hours. No significant difference between the time required to reach the desired pH was noted between the samples. The organoleptic properties of the yoghurt supplemented with plant sterols was poor. On storage,  
10 particles of plant sterol powder were found in the whey, on syneresis.

#### **Example 2.**

The yoghurt was made as described in Example 1 but plant sterols (1.7 gram) were added to whey proteins-fortified milk prior to  
15 homogenisation, the resulting suspension was homogenised, heated up to 55°C and left at this temperature for 2 hours. The yoghurt produced had a texture, appearance and flavour like the corresponding product composition prepared with no plant sterols added.

#### **20 Example 3.**

A 20 % by weight suspension of plant sterols was prepared by intensive homogenisation of finely divided sterol powder in water. 1 volume of 10% (w/w) sodium caseinate solution was added and after

stirring for 1 hour, the pH of the resulting suspension was adjusted to 4.5 with dilute HCl. The stirring was continued for 5 hours and the pH was adjusted to 6.4.

5   **Example 4.**

Plant sterols suspension was prepared as described in Example 3 but a 9% (w/w) solution of high protein whey powder was used instead of sodium caseinate. The resulting suspension was heated to 55°C and incubated at this temperature for 2 hours with stirring.

10

**Example 5.**

Plant sterols suspension was prepared as described in Example 3 but a 20% (w/w) solution of semi skimmed milk powder was used instead of sodium caseinate. The resulting suspension was incubated at room

15   temperature for 16 hours, with stirring.

**Example 6**

Plant sterol suspension was prepared as described in Example 3 but instead of pH adjustment, it was stirred for 1 hour and then spray-

20   dried.

**Example 7.**

Plant sterol suspension was prepared as described in Example 4 and spray-dried.

**5 Example 8.**

The yoghurt was made as described in Example 2 by admixing 1 volume of sterol suspension prepared as described in Examples 3, 4 and 5 with 10 volumes of milk prior to homogenisation. The yoghurts obtained had a similar texture, appearance and flavour to, or were substantially  
10 indistinguishable from, the corresponding product compositions which were made with no plant sterols added.

**Example 9.**

The yoghurt was made as described in Example 8 but using 75  
15 parts of soymilk (3.5% protein) fortified with high protein whey powder and 25 parts of pasteurised skimmed milk. The yoghurts obtained had a similar texture, appearance and flavour to, or were substantially indistinguishable from, the corresponding product compositions which were made with no plant sterols added.

20

**Example 10.**

The yoghurt base for Swiss style yoghurt was made as described in Example 8 but 0.75% of a standard premixed stabiliser containing

modified food starch, pectin and gelatine, and 3.5 % sugar was added. The stirred yoghurt was produced by breaking the coagulum and a standard fruit product (40-50% strawberry or raspberry; 30-40% sugar, 2-3% modified starch) was mixed in. The yoghurts obtained had a similar  
5 texture, appearance and flavour to, or were substantially indistinguishable from, the corresponding product compositions which were made with no plant sterols added.

#### **Example 11.**

10 Swiss style yoghurt was made as described in Example 10, but 2 volumes of sterol suspension prepared as described in Examples 3, 4 and 5 was mixed with 10 volumes of milk prior to homogenisation. The stirred yoghurt was produced by breaking the coagulum and a fruit product (40-50% strawberry or raspberry; 30-40% sugar, 2-3% modified starch) was  
15 mixed in. The yoghurts obtained had a texture, appearance and flavour like the product compositions which were made with no plant sterols added.

#### **Example 12.**

20 The yoghurt was made as described in Example 10 but 3 volumes of sterol suspension prepared as described in Examples 3, 4 and 5 was used. The yoghurts obtained had a similar texture and appearance to the corresponding product compositions which were made with no plant

sterols added but assessors commented on the sensation of powderiness in the mouth.

**Example 13.**

5           The yoghurt was made as described in Example 10 but 1.7 gram of plant sterols prepared as described in Examples 6 and 7 was added to milk prior to homogenisation. The yoghurts obtained had a similar texture, appearance and flavour to, or were substantially indistinguishable from, the corresponding product compositions which were made with no plant  
10   sterols added.

**Example 14.**

          The yoghurt was made as described in Example 13 but using stanol (>95% purity) and a technical grade preparation (approximately 50%  
15   stanol) which was purified by re-crystallisation from ethanol, instead of the plant sterol mixture of Example 1. Stanol was subjected to intensive homogenisation to reduce the crystal size, typically to about 50 micron. The yoghurts obtained had a texture and appearance like the corresponding product compositions which were made with no stanol  
20   added, although the assessors noted an aftertaste.

**Example 15.**

Stanol acetate was prepared by esterification of the stanol with acetic anhydride under standard conditions. The yoghurts were made as described in Examples 10 and 14. The yoghurts obtained had a texture, appearance and flavour like the corresponding product compositions which were made with no stanol acetate added.

**Example 16.**

A very low fat Swiss style yoghurt was prepared as described in Example 10 but using yoghurt base made from HTST skimmed milk (9% solids and less than 0.1% by weight butterfat) to which 0.75% of a standard premixed stabiliser containing modified food starch, pectin and gelatine, 6% of non heat non fat dry solids and 5 % sugar was added. After blending 1 part of fruit was added per 5 parts of yoghurt. The fat content of the yoghurt was about 0.2%.

**Example 17.**

The yoghurt was prepared as described in Example 10 but a standard Sundae style fruit (35-45% of strawberry or raspberry; 15-20% sugar 10-20% corn syrup, 2-4% modified starch and remaining % water) was placed on the bottom of the containers. The inoculated mixture of ingredients prepared as described in Example 10 was poured on top. Approximately 1 part of fruit product for 5 parts of yoghurt was used. The

products which were assessed after mixing the yoghurt with the fruit had a similar texture, appearance and flavour to, or were substantially indistinguishable from, the corresponding product compositions which were made with no plant sterols added.

5

**Example 18.**

A yoghurt base was prepared as described in Example 8. The product was cooled down to 15°C, and 3% of water and food grade citric acid was added to adjust the pH to about 4.0. 1.9% by weight of citric  
10 pectin and 4.1 % by weight of orange essence was added and, after intensive stirring, the mixture was homogenised under pressure and heated to about 50°C and cooled down to 3-5°C. The yoghurt drinks obtained had a similar texture, appearance and flavour to, or were substantially indistinguishable from, the corresponding product  
15 compositions which were made with no plant sterols added.

**Example 19.**

Yoghurt fruit was prepared by blending 35 parts strawberries, 5 parts water and 3 parts starch under constant agitation with heating. The  
20 resulting mixture was held at 89-95°C for about 10 minutes until it thickened and then 30 parts of 3:1 mixture of sugar and plant sterol prepared as described in Examples 6 and 7 was added with vigorous agitation together with additional 5 parts of sugar, 15 parts of corn syrup, 4



parts of natural flavouring, 1 part of preservative (potassium sorbate), 0.5 parts of colouring agent and 0.5 parts of citric acid. Vigorous agitation was continued on cooling.

5    **Example 20.**

Yoghurt fruit was prepared as described in Example 19 but finely divided plant sterol powder with a particle size around 30-50 micron or below, as assessed by microscopy, was used.

10   **Example 21.**

Yoghurt fruit was prepared as follows: Plant sterols prepared as described in Examples 6 and 7 were added to water and the resulting 20% (w/w) suspension was homogenised at 50°C. 12 parts of this suspension was then blended with 45 parts strawberries and 3 parts modified starch  
15   under constant agitation with heating. The resulting mixture was held at 89-95°C for about 10 minutes and then 40 parts of sugar and other ingredients as described in Example 19 were added with vigorous agitation which continued on cooling.

20   **Example 22.**

Yoghurts were made as described in Examples 11 and 17 with and without the addition of plant sterols but using the fruit products prepared as described in Examples 19, 20 and 21. The products which were

assessed after mixing the yoghurt with the fruit had a similar texture, appearance and flavour to, or were substantially indistinguishable from, the corresponding product compositions which were made with no plant sterols added.

5

**Example 23.**

Plant sterol suspension was prepared as described in Example 4 but Soya bean protein was used as a coating agent.

10 **Example 24.**

A 12 % by weight suspension of plant sterols was prepared by intensive homogenisation of finely divided sterol powder in cow's milk. The resulting suspension was pasteurised by heating up to 80°C, re-homogenised and used for the preparation of a beverage as described in

15 Example 30.

**Example 25.**

Sterol suspension was prepared as described in Example 24 but Soya milk was used instead of cow's milk.

20

**Example 26.**

A 10% suspension of coated lycopene was prepared as described in Examples 24 and 25 but using lycopene crystals instead of plant sterols.

**Example 27.**

Plant sterols (10 g) and lecithin (9.5 g) were dissolved in boiling ethanol (100 mL) and the resulting solution was added to 100 mL of water.

- 5 After ethanol was removed the coating of plant sterols was accomplished by adding 1 volume of 9% whey protein to 10 volumes of the sterol-lecithin suspension ( as described in Example 4).

**Example 28.**

- 10 Plant sterols (10 g) and lycopene-containing solids (effective concentration of lycopene 0.5 g) were dissolved in boiling ethanol (100 mL) and the resulting solution was added to 80 mL of water. After ethanol was removed the coating of plant sterols was accomplished by adding 1 volume of 9% whey protein to 10 volumes of the sterol-lycopene  
15 suspension.

**Example 29.**

- The yoghurt drink was made as described in Example 18 but 50 mg of soya isoflavons per helping was added to the product together with  
20 other prior to the homogenisation.

**Example 30.**

A milk shake was prepared to contain the sterol suspension prepared as described in Example 24 (15%) and non-fat dry milk (6%), whey concentrate (5%), sugar (9%) fructose-dextrose syrup (12%), corn  
5 syrup (3%), whey (5%), butter (7.2%), flavouring (0.5%), emulsifiers (0.3%) and water (37%). The syrups and flavouring were added to water combined with the sterol suspension and mixed under high sheer with heating up to the pasteurising temperature. Other components were added and, when the sugar dissolved, the mixture was homogenised and  
10 cooled directly into a heat-exchanger until the temperature of the product reached about 35°C. The product was stored overnight and then whipped.

**Example 31**

A milk shake was prepared as described in Example 30 but using  
15 the coated lycopene suspension (see Example 26) instead of coated plant sterols to give the effective concentration of 10 mg of lycopene per helping.

**Example 32**

20 A dough for bread was prepared to contain (in baker percentage): flour (100%), water and plant sterol suspension prepared as in Example 4 (66%), compressed yeast (2%), yeast food (0.5%), sucrose (4%), no-fat milk solids (4%), sucrose (4%), salts including sodium propionate (2.3%),

shortening and surfactants (2%). All ingredients were mixed in a Hobart mixer. The dough was proofed, scaled, hand-rounded and given a rest at room temperature. The rested dough was sheeted, moulded, placed in a lightly greased pans and proofed at high humidity. The bread was baked  
5 for 20 min at 215°C in a rotary rack oven. The amount of plant sterol used was sufficient to give 1.4% of plant sterols per dry weight of the final bread.

**Example 33.**

10 A suspension (18% w/w) of plant sterols was prepared by intensive homogenisation of finely divided sterol powder in 2% (w/w) solution of milk powder, heated to 60°C and maintained at this temperature for 30 minutes. Salt (5%) was added and 30 parts of the resulting mixture was emulsified with 70 parts of a mixture consisting of sunflower oil (79%) and  
15 hydrogenated vegetable oil (21%). Emulsifiers E471 and E322 (0.5%), fat soluble butter flavours (0.035%) and colouring (0.04%) were included in the oil.

**Example 34.**

20 A suspension (18% w/w) of plant sterols in water was prepared as described in Example 33 and 1 part of a 10% solution of whey powder was added to 5 parts of this suspension. After stirring for 30 minutes the resulting mixture was spray dried.

**Example 35.**

A fat spread was made as described in Example 33 but plant sterols prepared as described in Example 34 were used to make a suspension (18% w/w) of plant sterols in 5% salt solution which was used to prepare the aqueous phase of the spread.

**Example 36.**

A fat spread was made as described in Example 35 but the spray dried plant sterols were added to the oil phase (8.5% w/w). Substantial thickening was observed during homogenization which hampered processing. The texture and organoleptic properties of the product were inferior.

**Example 37.**

A fat spread was made as described in Example 33 but plant sterol suspension was prepared in buttermilk (6% w/w protein).

**Example 38.**

A suspension (18% w/w) of plant sterols in water was prepared by intensive homogenisation and 1 part of a 10% solution of whey powder was added to 5 parts of this suspension. The resulting mixture was heated to 60°C and maintained at this temperature for 90 minutes. Salt

(5%) was added and a fat spread was prepared as described in Example 33.

**Example 39.**

5           A fat spread was made as described in Example 38 but the oil phase of the spread contained 20% (w/w) of butterfat.

**Example 40.**

          A fat spread was made as described in Example 38 but the oil  
10       phase of the spread contained 7% of stanol esters which were prepared as described in US 5,502,045.

**Example 41.**

          A fat spread was prepared as described in Example 38 but 0.06%  
15        $\alpha$ -tocopherol (E307) was added to the oil phase and Soya bean isoflavones (0.1% w/v) were added to the aqueous phase prior to emulsification.

**Example 42.**

20       A suspension of plant sterols was prepared with whey powder as described in Example 35. The resulting mixture was homogenised, heated to 55°C and incubated at this temperature for 30 min. Gelatine (3%) and salt (2.5%) were added and pH was adjusted to 5.8 by addition

of citric acid. 62 parts of this suspension was emulsified with 38 parts of oil using E471 and E322 (1%) as emulsifiers.

**Example 43.**

5        A suspension (15% w/w) of plant sterols was prepared in an aqueous solution containing gelatine (3%) and salt (2.5%). The resulting mixture was homogenised, heated to 55°C and incubated at this temperature for 30 min. The suspension was emulsified with oil as described in Example 42. No satisfactory product was obtained as the  
10    emulsion gelled.

**Example 44.**

      A fat spread was made as described in Example 43 but using the plant sterols prepared as described in Example 34. Emulsification  
15    proceeded normally.

**Example 45.**

      380 mg of  $\beta$ -sitosterol was added to 5.0ml of sunflower oil containing either 1% lecithin or 0.4% E476. Both samples gave a clear  
20    solution at 60°C which gelled within 10-15 min if cooled down at room temperature or instantly on the addition of water. At the same concentration  $\beta$ -sitosterol was not soluble in sunflower oil in the absence of the emulsifiers. The experiment was repeated with 300 mg of stanol



and sunflower oil containing 1% lecithin (the same amount of stanol was not soluble in the oil in the absence of lecithin). The stanol/lecithin solution gelled on cooling or on the addition of water.

5   **Example 46.**

A suspension (18% w/w) of plant sterols was prepared by intensive homogenisation of finely divided sterol powder in water. 1 part of 10% (w/w) sodium caseinate solution was added per 5 parts of the sterol suspension and the pH of the resulting suspension was adjusted to 4.6  
10 with citric acid. The suspension was heated up to 90°C with stirring and incubated at this temperature for 1 h, then gelatine (3%), modified starch (3%) and salt (2%) were added and, after the added ingredients dissolved, the mixture was pasteurised at this temperature, cooled down to 50°C and used as an aqueous phase for the preparation of fat spread. The fat  
15 spread was prepared as described in Example 42 but using 71 parts and 29 parts of the above aqueous suspension and oil phase respectively.

**Example 47.**

A suspension of plant sterols was prepared as described in  
20 Example 46, and after stirring for 1 hour, spray-dried. An aqueous phase of fat spread was prepared to contain gelatine (3%), low DE maltodextrin (15%) and salt (2%) and citric acid (0.5%). Spray dried plant sterols (15%

w/w) were admixed into the aqueous solution above, the pH adjusted to 5.8 and a fat spread was made as described in Example 46.

**Example 48.**

- 5           A fat spread was made as described in Example 43 but using plant sterols prepared as described in Example 27. Emulsification proceeded normally.

**CLAIMS**

1. A method for making a composition suitable for inclusion in a food product or beverage the method comprising the step of combining a plant sterol, lycopene or other hydrophobic compound which can be shown to be beneficial for human health with a component which is acceptable as a food additive, wherein the component which is acceptable as a food additive interacts with the surface of the hydrophobic compound.
2. A method according to Claim 1 wherein the hydrophobic compound is any one of a plant sterol, lycopene (such as tomato lycopene), other carotenoids and poorly water soluble anti-oxidants or a combination thereof.
3. A method according to Claim 1 or 2 wherein the component which is acceptable as a food additive is any one of a food-acceptable polypeptide, polysaccharide or low molecular weight substance with similar chemical functionality/functional groups.
4. A method according to any one of Claims 1 to 3 wherein the component acceptable as a food additive is a coating agent which is encouraged to adhere to the surface of the plant sterol, lycopene or other hydrophobic compound.

5. A method according to any one of Claims 1 to 4 wherein the plant sterol, lycopene or other hydrophobic compound is contacted with the component acceptable as a food additive in aqueous suspension.

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6. A method according to any one of Claims 1 to 5 which comprises coating the plant sterol, lycopene or other hydrophobic compound with a food additive by combining the hydrophobic compound and the food additive in an aqueous suspension and incubating the resulting mixture, with stirring, at a temperature between 18°C and 50°C.

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7. A method as claimed in Claim 6, where the suspension containing the plant sterol, lycopene or other hydrophobic compound, and a food additive is heat-treated at a temperature above 50°C.

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8. A method as claimed in any of the Claims 6 and 7, where the pH of the suspension containing the plant sterol, lycopene or other hydrophobic compound, and a food additive is adjusted to substantially neutralise the net charge on the molecule of the food additive and, preferably close to the isoelectric point of the food additive.

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9. A method as claimed in any one of Claims 6 to 8, where the coated plant sterol, lycopene, or other hydrophobic compound, is recovered in dry form.
- 5 10. A method as claimed in any of the Claims 1 to 9, where the food additive is a multifunctional substance and contains at least one carboxyl group and at least one hydroxyl group.
- 10 11. A method as claimed in any of the Claims 1 to 9, where the food additive is a protein.
12. A method as claimed in any of the claims 1 to 9, where the food additive is derived from milk.
- 15 13. A method as claimed in Claim 12, where the food additive is selected from the group comprising casein, caseinate, whey protein, whey, milk powder, buttermilk and butterfat.
- 20 14. A method as claimed in any of the Claims 1 to 13, where the hydrophobic compound is a plant sterol selected from the group comprising  $\beta$ -sitosterol,  $\beta$ -campesterol,  $\beta$ -stigmasterol,  $\beta$ -sitostanol,  $\beta$ -campestanol and  $\beta$ -stigmastanol and their carboxylic acid esters, and mixtures thereof.

15. A method according to any one of Claims 1 to 14 wherein lecithin is additionally combined with the plant sterol, lycopene or other hydrophobic compound and the component which is acceptable as a food additive.

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16. A method according to Claim 15 wherein the lecithin and hydrophobic compound are combined prior to combination with the component which is acceptable as a food additive.

10 17. A method according to Claim 15 or 16 wherein there is more hydrophobic compound than lecithin by weight.

18. A method according to any one of Claims 15 to 17 where the hydrophobic compound and lecithin are combined by dissolving them in a  
15 common solvent and then precipitated prior to combining them with a component which is acceptable as a food additive.

19. A method according to any one of Claims 1 to 17 wherein the composition is formed *in situ* in the preparation of the food product or  
20 beverage.

20. A composition suitable for inclusion in a food product or beverage obtainable by the method of any one of Claims 1 to 18.

21. A composition suitable for inclusion in a food product or beverage comprising a plant sterol, lycopene or other hydrophobic compound which can be shown to be beneficial for human health with a component which is acceptable as a food additive coated on the surface thereof.

22. A composition according to Claim 20 or 21 wherein the hydrophobic compound is a plant sterol, lycopene or other carotenoid and poorly water soluble anti-oxidants or a combination thereof.

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23. A composition according to any one of Claims 20 to 22 wherein the component acceptable as a food additive is a food-acceptable polypeptide, polysaccharide or low molecular weight substance with similar chemical functionality/functional groups.

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24. A dry ingredient which is obtained by drying, preferably spray-drying of a mixture containing a milk derived solid and plant sterols, where the milk derived solid and the plant sterols are combined in a process wherein the milk derived solid and the plant sterols are contacted in an aqueous suspension, and where the milk derived solid is selected from the group consisting of casein, caseinate, whey protein, whey, milk powder, buttermilk and butterfat, and where the plant sterols are selected from the group comprising  $\beta$ -sitosterol,  $\beta$ -campesterol,  $\beta$ -stigmasterol,  $\beta$ -sitostanol,

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$\beta$ -campestanol and  $\beta$ -stigmastanol and their carboxylic acid esters, and a mixture thereof.

25. A dry ingredient according to Claim 24 wherein the contact is  
5 carried out at a temperature above 40°C and preferably with stirring.

26. A dry ingredient according to Claim 24 or 25 wherein the contact is carried out at a temperature and exposure time combination which is sufficient for pasteurisation.

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27. A composition suitable for inclusion in a food product or beverage consisting essentially of a plant sterol, lycopene or other hydrophobic compound which can be shown to be beneficial to human health and lecithin wherein there is more hydrophobic compound than lecithin by  
15 weight.

28. A composition suitable for inclusion in a food product or beverage according to Claim 27 wherein the plant sterol, lycopene or other hydrophobic compound is coated with a component which is acceptable  
20 as a food additive.

29. A method for preparing a food product or beverage which is supplemented with a plant sterol, lycopene or other hydrophobic compound



which can be shown to be beneficial for human health the method comprising the step of (1) carrying out the method of any one of Claims 1 to 17 and 19 *in situ* during the preparation process or (2) adding at an appropriate stage during the preparation process a composition according to any one of Claims 20 to 28 or (3) a combination of (1) and (2).

30. A food product or beverage which is supplemented with a plant sterol, lycopene or other hydrophobic compound which can be shown to be beneficial for human health obtainable by the method of Claim 29.

31. A food product or beverage according to Claim 30 which is any one of a fermented or non-fermented dairy product, an emulsified fat product, a non-dairy product, alcoholic and non-alcoholic drinks, soups, sauces, dips, salad dressings, mayonnaise, non-fat spreads, confectionery, bread, cakes, biscuits, and breakfast cereals.

32. A food product or beverage according to Claim 30 which is produced by the fermentation of a mixture consisting of a fermentable raw material and plant sterols with lactic acid bacteria, and which has a similar texture, appearance and flavour to, or is substantially indistinguishable from, the corresponding product composition which is made with no plant sterols added.

33. A product as claimed in Claim 32, where the fermented food product is a fermented milk product.

34. A product as claimed in Claim 33, where the fermented milk product is selected from the group comprising of yoghurt, yoghurt product, kefir, ymer, buttermilk, butterfat, sour cream, sour whipped cream and fresh cheese.

35. A product as claimed in any of the Claims 32 to 34, where the fermentation is carried out with more than one micro-organism.

36. A product as claimed in any of the Claims 32 to 35, where the micro-organism is selected from the group comprising *Lactobacillus sp.*, *Streptococcus sp.*, *Lactococcus sp.*, *Bifidobacterium sp.*, and mixtures thereof.

37. A product as claimed in any of the Claims 32 to 36, which contains less than 2% fat.

38. A product as claimed in any of the Claims 32 to 37, where plant sterols are selected from the group comprising  $\beta$ -sitosterol,  $\beta$ -campesterol,  $\beta$ -stigmasterol,  $\beta$ -sitostanol,  $\beta$ -campestanol and  $\beta$ -stigmastanol and their carboxylic acid esters, and mixtures thereof.

39. A food product which is produced by the fermentation of a mixture consisting of a fermentable raw material and plant sterols with lactic acid bacteria, and which has a similar texture, appearance and flavour to, or is  
5 substantially indistinguishable from, the corresponding product composition which is made with no plant sterols added.

40. A process for the preparation of the products as claimed in Claim 39, where plant sterols are added to a fermentable raw material prior to  
10 inoculation with micro-organisms.

41. A process for the preparation of the products as claimed in Claim 39, where plant sterols are added to a fermentable raw material prior to pasteurisation.

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42. A process for the preparation of the products as claimed in Claim 39, where the mixture of a fermentable raw material and plant sterols is heat treated at a temperature above 50°C prior to inoculation with a micro-organism.

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43. A process as claimed in any of the Claims 40 to 42, where the raw material is milk or is derived from milk.

44. A process as claimed in any of the Claims 40 to 43 where the plant sterols used are coated with food additives as claimed in any of the Claims 6 to 18.

5 45. A product as claimed in any of the Claims 32 to 38, which is prepared in the process as claimed in any of the Claims 40 to 43 where the plant sterols used are coated with food additives as claimed in any of the Claims 6 to 18.

10 46. An emulsified fat spread according to Claim 30 which is supplemented with plant sterols, where the plant sterols are included into the aqueous phase of the product composition in a largely insoluble form.

15 47. An emulsified fat spread as claimed in Claim 46, which contains milk derived solids.

48. An emulsified fat spread as claimed in any of the Claims 46 to 47, where milk derived solids are selected from the group consisting of casein, caseinate, whey protein, whey, milk powder, buttermilk and butterfat.  
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49. An emulsified fat spread as claimed in any of the Claims 46 to 48, which contains at least one additional dietary supplement in addition to those required by legislation.

50. An emulsified fat spread as claimed in any of the Claims 46 to 49, which contains less than 40% fat.

5 51. An emulsified fat spread as claimed in any of the Claims 46 to 50, where the fat phase is supplemented with oil-soluble plant sterols.

52. An emulsified fat spread as claimed in any of the Claims 46 to 51, where the fat soluble plant sterols are different from the plant sterols  
10 included in the aqueous phase.

53. An emulsified fat spread as claimed in any of the Claims 46 to 52, where the plant sterols are selected from the group comprising  $\beta$ -sitosterol,  $\beta$ -campesterol,  $\beta$ -stigmasterol,  $\beta$ -sitostanol,  $\beta$ -campestanol and  
15  $\beta$ -stigmastanol and their carboxylic acid esters, and a mixture thereof.

54. An emulsified fat spread which is supplemented with plant sterols, where the plant sterols are included into the aqueous phase of the product composition in a largely insoluble form.

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55. A process for the production of products as claimed in Claim 54, comprising of contacting milk derived solids and plant sterols in the aqueous phase of the product composition prior to emulsification.

56. A process as claimed in Claim 55, where the contact is carried out at a temperature above 40°C and, preferably with stirring.

5 57. A process as claimed in any of the Claims 55 and 56, where the contact is carried out at a temperature and exposure time combination which is sufficient for pasteurisation.

58. A yoghurt which is prepared by (i) admixing milk and milk derived  
10 solids with plant sterols, and heating the mixture to a temperature above 60°C, and (ii) cooling the mixture down, and (iii) adding a starter culture containing micro-organisms selected from the group comprising *Lactobacillus sp.*, *Streptococcus sp.*, *Lactococcus sp.* and *Bifidobacterium sp.*, and mixtures thereof and (iv) incubating the resulting mixture until the  
15 milk coagulates.

59. A yoghurt as claimed in Claim 58, which is prepared with non-milk derived ingredients selected from the group comprising fruit products, stabilisers, thickeners, sweeteners, flavouring and colouring agents, and a  
20 mixture thereof.

60. A yoghurt as claimed in any of the Claims 58 and 59, which contains less than 2% fat.

61. A yoghurt as claimed in any of the Claims 58 to 60, where the plant sterols used are coated with food additives as claimed in any of the Claims 6 to 18.

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62. A yoghurt product prepared from the yoghurt as claimed in any of the Claims 58 to 61.

63. A process for the preparation of stirred yoghurt and yoghurt products which contain (i) plant sterols and (ii) non-milk derived ingredients selected from the group comprising fruit products, stabilisers, thickeners, sweeteners, flavouring and colouring agents, and mixtures thereof, by admixing the plant sterol into the yoghurt or the yoghurt product during or after breaking the coagulum.

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64. A yoghurt and a yoghurt product which are prepared by the process as claimed in Claim 63, where the plant sterols are prepared by the method as claimed in any of the Claims 6 to 18.

20 65. A process for the preparation of yoghurt fruit containing plant sterols which comprises (i) blending a mixture of fruit and water and plant sterols and ingredients selected from the group consisting of stabilisers, thickeners, sweeteners, flavouring and colouring agents and preservatives

and (ii) heating up the mixture to a temperature sufficient to pasteurise the mixture and (iii) holding the mixture at elevated temperature until it thickens and (iv) cooling the mixture down, with agitation.

- 5 66. A process as claimed in Claim 65, where plant sterols used are prepared by the method as claimed in any of the Claims 6 to 18.

67. A yoghurt composition which consists of three essential ingredients: (i) yoghurt base; (ii) yoghurt fruit and (iii) free (unesterified) plant sterols.

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68. A yoghurt composition which consists of four essential ingredients: (i) yoghurt base; (ii) yoghurt fruit, (iii) free (unesterified) plant sterols and (iv) a non-milk derived ingredient selected from the group comprising stabilisers, thickeners, sweeteners, flavouring and colouring agents, and  
15 mixtures thereof.

69. A yoghurt composition as claimed in any of the Claims 67 and 68, where plant sterols used are coated with food additives as claimed in any of the Claims 6 to 18.

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70. A yoghurt composition as claimed in any of the Claims 67 and 68, where the yoghurt fruit is prepared as claimed in any of the Claims 59 and 60.



71. A yoghurt composition as claimed in any of the Claims 67 to 70, where plant sterols are incorporated both into the yoghurt base and the yoghurt fruit.

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72. A yoghurt composition as claimed in any of the Claims 67 to 71, which contains less than 2% fat.

73. A yoghurt, a yoghurt product and a yoghurt composition as claimed  
10 in any of the Claims 58 to 62, 63 and 67 to 72, where plant sterols are selected from the group comprising  $\beta$ -sitosterol,  $\beta$ -campesterol,  $\beta$ -stigmasterol,  $\beta$ -sitostanol,  $\beta$ -campestanol and  $\beta$ -stigmastanol and mixtures thereof.

15 74. An organogel composition consisting of (i) a liquid fat component, (ii) at least one emulsifying agent and (iii) at least one sterol selected from the group comprising  $\beta$ -sitosterol,  $\beta$ -campesterol,  $\beta$ -stigmasterol,  $\beta$ -sitostanol,  $\beta$ -campestanol and  $\beta$ -stigmastanol.

20 75. An organogel composition as claimed in Claim 74, where the emulsifying agent is selected from the group comprising lecithin and polyglycerol esters and mixtures thereof.

76. The use of the organogel compositions as claimed in any of the Claims 74 and 75 in food and cosmetic products.

77. The use of a dry ingredient as claimed in Claim 24 in food and  
5 pharmaceutical compositions.

78. The use of a composition according to any one of Claims 20 to 28 in food, food products or beverages.

10 79. The use according to Claim 78 wherein the food is an emulsified fat spread.

80. A method, product or use according to any one of the preceding claims wherein a further additive which can be shown to be beneficial to  
15 human health is used.

81. A method, product or use according to Claim 80 wherein the further additive is a further hydrophobic compound which can be shown to be beneficial for human health, such as a different plant sterol or lycopene.

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82. A method, product or use according to Claim 80 or 81 wherein the further additive is soluble in aqueous phase of food product or beverage.

83. A method, product or use according to Claim 81 wherein the further additive is a soya bean isoflavone.

